Requester's Full Name: Khatol Shahmn-shah Examiner #: 78526 Date: 8/28/01
Art Unit: 1645 Phone Number 308-8896 Serial Number: 09/840,458
Mail Box and Bldg/Room Location: 8D-17 Results Format Preferred (circle): PAPER DISK E-MAIL
8E-12

If more than one search is submitted, please prioritize searches in order of need.

****

Please provide a detailed statement of the search topic, and describe as specifically as possible the subject matter to be searched. Include the elected species or structures, keywords, synonyms, acronyms, and registry numbers, and combine with the concept or utility of the invention. Define any terms that may have a special meaning. Give examples or relevant citations, authors, etc, if known. Please attach a copy of the cover sheet, pertinent claims, and abstract.

Title of Invention: _____

Inventors (please provide full names): See attached Bib Sheet

Earliest Priority Filing Date: 4/25/2000

***For Sequence Searches Only* Please include all pertinent information (parent, child, divisional, or issued patent numbers) along with the appropriate serial number.**

Please search claims 1-25 attached claims and abstracts

Full search Including authors and meetings

Thanks Khatol

Point of Contact:
Mary Hale
Technical Info. Specialist
CM1 12D16 Tel: 308-4258

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****

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Searcher Location: _____
Date Searcher Picked Up: _____
Date Completed: 8/30
Searcher Prep & Review Time: _____

Type of Search

NA Sequence (#) _____ STN 106,160
AA Sequence (#) _____ Dialog _____
Structure (#) _____ Questel/Orbit _____
Bibliographic ✓ Dr.Link _____
Litigation _____ Lexis/Nexis _____
Fulltext _____ Sequence Systems _____

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K. S-Shah
840485

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FILE COVERS 1947 - 30 Aug 2001 VOL 135 ISS 10
FILE LAST UPDATED: 29 Aug 2001 (20010829/ED)

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E#	FREQUENCY	AT	TERM
E1	0	1	MEROTERPENOID/CT
E2	0	3	MEROTERPENOID QUINONES/CT
E3	0	2 -->	MEROZOITE/CT
E4	0	2	MEROZOITE SURFACE PROTEIN 1/CT
E5	0	1	MERPIQUAT/CT

```
=> e e3+all/ct
E1            0   -->  Merozoite/CT
E2                 USE  Development, microbial (L) merozoite/CT
*********  END***

=> e sarcocystis neurona/ct 5
```

```
E#   FREQUENCY    AT     TERM
--   ---------    --     ----
E1           1     5     SARCOCYSTIS MUCOSA/CT
E2          18     5     SARCOCYSTIS MURIS/CT
E3          31     5 --> SARCOCYSTIS NEURONA/CT
E4           1           SARCOCYSTIS OVICANIS/CT
E5           3     5     SARCOCYSTIS OVIFELIS/CT

=> e e3+all/ct
E1        2314   BT4  Eukaryote (Eukaryotae)/CT
E2          75     BT3  Apicomplexa/CT
E3          74       BT2  Coccidia/CT
E4          46         BT1  Sarcocystis/CT
E5          31           -->  Sarcocystis neurona/CT
                            HN   Valid heading during volume 116 (1992) to
                                 present.
*********  END***

=> s e4-5
            46 SARCOCYSTIS/CT
            31 "SARCOCYSTIS NEURONA"/CT
L1          71 (SARCOCYSTIS/CT OR "SARCOCYSTIS NEURONA"/CT)

=> e tachyzoite/ct 5
E#   FREQUENCY    AT     TERM
--   ---------    --     ----
E1           3    11     TACHYSURUS THALASSINUS/CT
E2           2    11     TACHYSURUS ZONA/CT
E3           0       --> TACHYZOITE/CT
E4           1           TACITUS BELLUS/CT
E5           0     1     TACKACE/CT

=> e neospora hughesi/ct
E#   FREQUENCY    AT     TERM
--   ---------    --     ----
E1          30     6     NEOSPORA/CT
E2          97     5     NEOSPORA CANINUM/CT
E3           6     5 --> NEOSPORA HUGHESI/CT
E4           0     8     NEOSTENANTHERA/CT
E5           1     8     NEOSTENANTHERA GABONENSIS/CT
E6           0    13     NEOSTIGMINE/CT
E7           0     8     NEOSTREARIA/CT
E8           1     8     NEOSTREARIA FLECKERI/CT
E9         368     2     NEOSTRIATUM/CT
E10          0     2     NEOSTRIATUM (L) DOPAMINERGIC SYSTEM/CT
E11          0     2     NEOSTRIATUM BRAIN/CT
E12          0     2     NEOSTRIATUM, DOPAMINERGIC SYSTEM BRAIN/CT

=> e e3+all/ct
E1        2314   BT4  Eukaryote (Eukaryotae)/CT
E2          75     BT3  Apicomplexa/CT
E3          74       BT2  Coccidia/CT
E4          30         BT1  Neospora/CT
E5           6           -->  Neospora hughesi/CT
*********  END***
```


Prepared by M. Hale 308-4258

```
=> s e5
L2            6 "NEOSPORA HUGHESI"/CT

=> e protozocidal/ct 5
E#   FREQUENCY     AT      TERM
--   ---------     --      ----
E1           0      2      PROTOZOAL INFECTION (L) SLEEPING SICKNESS/CT
E2           0      2      PROTOZOAN/CT
E3           0        --> PROTOZOCIDAL/CT
E4           0     12      PROTRACHEONISCUS/CT
E5           2     12      PROTRACHEONISCUS ORIENTALIS/CT

=> e epm/ct 5
E#   FREQUENCY     AT      TERM
--   ---------     --      ----
E1           0      2      EPK/CT
E2           0      1      EPL/CT
E3           0      1 --> EPM/CT
E4           0      2      EPM 4050/CT
E5           0      2      EPM 4060N/CT

=> e e3+all/ct
E1           0    -->  EPM/CT
*********  END***

=> e equine protozoal myeloencephal?/ct 5
E#   FREQUENCY     AT      TERM
--   ---------     --      ----
E1           0      2      EQUINE MORBILLIVIRUS/CT
E2           2      7      EQUINE PAPILLOMAVIRUS/CT
E3           0        --> EQUINE PROTOZOAL MYELOENCEPHAL?/CT
E4           2      6      EQUINE RHINOVIRUS/CT
E5           1      6      EQUINE RHINOVIRUS 1/CT

=> e immune response/ct 5
E#   FREQUENCY     AT      TERM
--   ---------     --      ----
E1           0     31      IMMUNE PROCESSES AND PHENOMENA (NON-CA HEADING)/CT
E2           0     65      IMMUNE RECEPTORS (NON-CA HEADING)/CT
E3           0      2 --> IMMUNE RESPONSE/CT
E4           0      2      IMMUNE SERUMS/CT
E5           0      2      IMMUNE SUPPRESSANTS/CT

=> e e3+all/ct
E1           0    -->  Immune response/CT
E2       23219      USE  Immunity/CT
*********  END***

=> s e2
L3       23219 IMMUNITY/CT

=> e equine dermal/ct 5
E#   FREQUENCY     AT      TERM
--   ---------     --      ----
```

```
E1            0      2      EQUINE CHORIONIC GONADOTROPIN/CT
E2            1             EQUINE CORONAVIRUS/CT
E3            0        --> EQUINE DERMAL/CT
E4            1      5      EQUINE FOAMY VIRUS/CT
E5            0      2      EQUINE HERPES 5 VIRUS/CT

=> e maiden darby bovine/ct
E#   FREQUENCY    AT      TERM
--   ---------    --      ----
E1            1      6      MAIASAURA PEEBLESORUM/CT
E2            0      1      MAICHUON/CT
E3            0        --> MAIDEN DARBY BOVINE/CT
E4            0      1      MAIDENHAIR/CT
E5            0      2      MAIDENHAIR TREE/CT
E6            0      1      MAIDENI/CT
E7            0      1      MAIDENII/CT
E8            0      1      MAIDIDUS/CT
E9            0      1      MAIDIRADICIS/CT
E10           0      1      MAIDIS/CT
E11           0      2      MAIDISM/CT
E12           0      2      MAIDS/CT

=> fil medli
COST IN U.S. DOLLARS                                SINCE FILE      TOTAL
                                                       ENTRY    SESSION
FULL ESTIMATED COST                                     8.17     432.50

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)          SINCE FILE      TOTAL
                                                       ENTRY    SESSION
CA SUBSCRIBER PRICE                                     0.00     -26.46

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 FILE LAST UPDATED: 28 AUG 2001 (20010828/UP).  FILE COVERS 1958 TO DATE.

 On April 22, 2001, MEDLINE was reloaded.  See HELP RLOAD for details.

 MEDLINE now contains IN-PROCESS records.  See HELP CONTENT for details.

 MEDLINE is now updated 4 times per week.  A new current-awareness alert
 frequency (EVERYUPDATE) is available.  See HELP UPDATE for more information.

 MEDLINE thesauri in the /CN, /CT, and /MN fields incorporate the
 MeSH 2001 vocabulary.  Enter HELP THESAURUS for details.

 The OLDMEDLINE file segment now contains data from 1958 through 1965.
 Enter HELP CONTENT for details.

 Left, right, and simultaneous left and right truncation are available in the
 Basic Index.  See HELP SFIELDS for details.

 THIS FILE CONTAINS CAS REGISTRY NUMBERS FOR EASY AND ACCURATE
 SUBSTANCE IDENTIFICATION.

=> e maiden darby bovine/ct
```

```
E#   FREQUENCY    AT     TERM
--   ---------    --     ----
E1           0     2     MAI INFECT/CT
E2           0     2     MAIC/CT
E3           0       --> MAIDEN DARBY BOVINE/CT
E4           0     1     MAIDENHAIR/CT
E5           0     2     MAIDENHAIR TREE/CT
E6           0     2     MAIDS/CT
E7           0     2     MAIL/CT
E8           0     2     MAIL DISTRIBUTION/CT
E9           0     2     MAIL DISTRIBUTIONS/CT
E10          0     2     MAIL ORDER/CT
E11          0     2     MAIL, ELECTRONIC/CT
E12          0     2     MAIL-ORDER/CT

=> e equine dermal/ct 5
E#   FREQUENCY    AT     TERM
--   ---------    --     ----
E1           0     2     EQUINE ARTERITIS VIRUSES/CT
E2           0     2     EQUINE COITAL EXANTHEMA VIRUS/CT
E3           0       --> EQUINE DERMAL/CT
E4           0     2     EQUINE DISEASE/CT
E5           0     2     EQUINE DISEASES/CT

=> e  canine monocyte/ct 5
E#   FREQUENCY    AT     TERM
--   ---------    --     ----
E1           0     2     CANINE INFECTIOUS HEPATITIS/CT
E2           0     2     CANINE INFECTIOUS HEPATITIS VIRUS/CT
E3           0       --> CANINE MONOCYTE/CT
E4           0     2     CANINE PARVOVIRUS/CT
E5           0     2     CANINE PARVOVIRUSES/CT

=> e mouse monocyte/ct 5
E#   FREQUENCY    AT     TERM
--   ---------    --     ----
E1           0     2     MOUSE LYMPHOCYTE PROTEIN MOIETY REDUCED OF
INTERFERON
                         TYPE II/CT
E2           0     2     MOUSE MAMMARY TUMOR VIRUS/CT
E3           0       --> MOUSE MONOCYTE/CT
E4           0     2     MOUSE MUTANT STRAIN/CT
E5           0     2     MOUSE MUTANT STRAINS/CT

=> e fetal rhesus monkey kidney/ct 5
E#   FREQUENCY    AT     TERM
--   ---------    --     ----
E1          40            FETAL RESORPTION: VE, VETERINARY/CT
E2           0     2     FETAL RESORPTIONS/CT
E3           0       --> FETAL RHESUS MONKEY KIDNEY/CT
E4           0     2     FETAL STRUCTURE/CT
E5           0     2     FETAL STRUCTURES/CT

=> e feline kidney/ct 5
E#   FREQUENCY    AT     TERM
```

```
--   ---------    --      ----
E1           7            FELINE INFECTIOUS PERITONITIS: TM, TRANSMISSION/CT
E2          16            FELINE INFECTIOUS PERITONITIS: VI, VIROLOGY/CT
E3           0        --> FELINE KIDNEY/CT
E4           0     2      FELINE LENTIVIRUS/CT
E5           0     2      FELINE LENTIVIRUSES/CT

=> fil medl,caplus,biosis,embase,wpids,jicst
COST IN U.S. DOLLARS                                SINCE FILE      TOTAL
                                                         ENTRY    SESSION
FULL ESTIMATED COST                                       0.60     433.10

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)          SINCE FILE      TOTAL
                                                         ENTRY    SESSION
CA SUBSCRIBER PRICE                                       0.00     -26.46

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=> s (l1 or l2 or merozoite? or sarcocystis neurona or tachyzoite or neospor?
hughes? or protozocid?)
L4         3707 FILE MEDLINE
L5         1804 FILE CAPLUS
L6         3473 FILE BIOSIS
L7         2743 FILE EMBASE
L8          108 FILE WPIDS
L9          179 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L10       12014 (L1 OR L2 OR MEROZOITE? OR SARCOCYSTIS NEURONA OR TACHYZOITE
OR
                NEOSPOR? HUGHES? OR PROTOZOCID?)

=> s (epm or equine protozoa? myeloencephal?)
L11         376 FILE MEDLINE
L12         856 FILE CAPLUS
L13         472 FILE BIOSIS
L14         332 FILE EMBASE
L15         380 FILE WPIDS
```

L16 75 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L17 2491 (EPM OR EQUINE PROTOZOA? MYELOENCEPHAL?)

=> s (equine derma? or maiden darby (w)(canine or bovine)(w)kidney or african green monkey kidney or (canine or mouse)(w)monocyte or fetal rhesus monkey kidney or feline kidney or baby hamster kidney)(2a)cell!
L18 1449 FILE MEDLINE
L19 1645 FILE CAPLUS
L20 6592 FILE BIOSIS
L21 1282 FILE EMBASE
L22 75 FILE WPIDS
L23 39 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L24 11082 (EQUINE DERMA? OR MAIDEN DARBY (W)(CANINE OR BOVINE)(W) KIDNEY OR AFRICAN GREEN MONKEY KIDNEY OR (CANINE OR MOUSE)(W) MONOCYTE
OR FETAL RHESUS MONKEY KIDNEY OR FELINE KIDNEY OR BABY HAMSTER KIDNEY)(2A) CELL!

=> s l10 and l17 and l24
L25 1 FILE MEDLINE
L26 0 FILE CAPLUS
L27 1 FILE BIOSIS
L28 1 FILE EMBASE
L29 1 FILE WPIDS
L30 0 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L31 4 L10 AND L17 AND L24

=> dup rem l31
PROCESSING COMPLETED FOR L31
L32 2 DUP REM L31 (2 DUPLICATES REMOVED)

=> d cbib abs 1-2

L32 ANSWER 1 OF 2 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD
AN 2000-571969 [53] WPIDS
CR 2001-218486 [22]
AB WO 200049049 A UPAB: 20010421
NOVELTY - Detection of **Sarcocystis neurona** in horses by identifying a specific antibody-antigen complex via an immunoassay is new.

DETAILED DESCRIPTION - Detection of **Sarcocystis neurona** in an equine in an immunoassay is improved by reacting a biological sample from the horse suspected of harboring the S. neurona with an antibody (Ab) which is selective in binding to an identifying S. neurona antigen (Ag) to form an Ab-Ag complex.

INDEPENDENT CLAIMS are also included for the following:

(1) a kit for detecting S. neurona in a biological sample from an equine;

(2) monoclonal antibodies against 16 plus or minus 4 kDa or 30 plus

or minus 4 kDa antigens of S. neurona; and
(3) isolated DNA sequences encoding the 16 plus or minus 4 kDa and
30
plus or minus 4 kDa antigens of S. neurona.
USE - The methods and antibodies are useful for detecting S. neurona (claimed) which causes **equine protozoal myeloencephalitis**, a neurological disorder in horses.
Dwg.0/0

L32 ANSWER 2 OF 2 MEDLINE DUPLICATE 1
2000043702 Document Number: 20043702. PubMed ID: 10577742. Simplified technique for isolation, excystation, and culture of Sarcocystis species from opossums. Murphy A J; Mansfield L S. (Animal Health Diagnostic Laboratory, Michigan State University, East Lansing 48824, USA.) JOURNAL OF PARASITOLOGY, (1999 Oct) 85 (5) 979-81. Journal code: JL3; 7803124. ISSN: 0022-3395. Pub. country: United States. Language: English.
AB **Sarcocystis neurona** is a protozoan parasite that causes a neurological disease in horses called **equine protozoal myeloencephalitis**. The route of transmission is speculated to be by fecal-oral transfer of sporocysts shed from opossums. Controversy exists regarding both the natural life cycle for this parasite as well as the species identity of opossum Sarcocystis. To provide stage-specific material for species comparison, 27 opossums from southern Michigan were screened for Sarcocystis spp. sporocysts. Seven opossums were positive for Sarcocystis sporocysts by fecal flotation. A simplified, effective technique for isolation, excystation, and culture
of
opossum Sarcocystis sp. from mucosal scrapings was developed. All 7 Sarcocystis sp. isolates were successfully cultured to grow long term in **equine dermal cells** to the **merozoite** stage. **Merozoites** were observed between 5 and 15 days after inoculation. In conclusion, opossums shed Sarcocystis sp. sporocysts that may be manipulated to excyst and grow in vitro in equine dermal cell
lines
to the **merozoite** stage using the simplified technique described.

=> s l10 and l17 and (l3 or immunity or immune response or immunogen?)
L33 5 FILE MEDLINE
L34 3 FILE CAPLUS
L35 3 FILE BIOSIS
L36 4 FILE EMBASE
L37 1 FILE WPIDS
L38 0 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L39 16 L10 AND L17 AND (L3 OR IMMUNITY OR IMMUNE RESPONSE OR IMMUNOGEN?
)

=> s l39 not l31
L40 5 FILE MEDLINE
L41 3 FILE CAPLUS
L42 3 FILE BIOSIS
L43 4 FILE EMBASE

L44 1 FILE WPIDS
L45 0 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L46 16 L39 NOT L31

=> dup rem l46
PROCESSING COMPLETED FOR L46
L47 8 DUP REM L46 (8 DUPLICATES REMOVED)

=> d cbib abs 1-8

L47 ANSWER 1 OF 8 CAPLUS COPYRIGHT 2001 ACS
2001:167817 Document No. 134:221431 Vaccine to control **equine protozoal myeloencephalitis** in horses. Mansfield, Linda S.; Rossano, Mary G.; Murphy, Alice J.; Vrable, Ruth A. (Michigan State University, USA). PCT Int. Appl. WO 2001015708 A1 20010308, 57 pp. DESIGNATED STATES: W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM; RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, CY, DE, DK, ES, FI, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG. (English). CODEN: PIXXD2. APPLICATION: WO 2000-US24221 20000831. PRIORITY: US 1999-PV152193 19990902; US 2000-513086 20000224.
AB The present invention provides vaccines and methods for making the vaccines that actively or passively protect an equid or other animal against **Sarcocystis neurona**. In particular, the present invention provides vaccines that provide active **immunity** which comprise a polypeptide or DNA vaccine that contains or expresses at least one epitope of an antigen that has an amino acid sequence substantially similar to a unique 16 (+/-4) kDa antigen and/or 30 (+/-4) kDa antigen of **Sarcocystis neurona**. The present invention further provides a vaccine that provides passive **immunity** to **Sarcocystis neurona** comprising polyclonal or monoclonal antibodies against at least one epitope of an antigen substantially similar to a unique 16 (+/-4) kDa antigen and/or 30 (+/-4) kDa antigen of **Sarcocystis neurona**.

L47 ANSWER 2 OF 8 MEDLINE DUPLICATE 1
2001434376 Document Number: 21125368. PubMed ID: 11226451. Molecular comparison of the dense granule proteins GRA6 and GRA7 of **Neospora hughesi** and Neospora caninum. Walsh C P; Vemulapalli R; Sriranganathan N; Zajac A M; Jenkins M C; Lindsay D S. (Center for Molecular Medicine and Infectious Diseases, Department of Biomedical Sciences and Pathobiology, Virginia-Maryland Regional College of Veterinary Medicine, Blacksburg, VA 24061-0342, USA.) INTERNATIONAL JOURNAL FOR PARASITOLOGY, (2001 Mar) 31 (3) 253-8. Journal code: GSB; 0314024. ISSN: 0020-7519. Pub. country: England: United Kingdom.
Language:
English.
AB **Neospora hughesi** is a recently described apicomplexan parasite that has been associated with several cases of **equine**

protozoal myeloencephalitis. The biology of this new parasite is just beginning to be defined. Towards this understanding, we report important differences between the nucleotide and deduced amino acid sequences of the dense granule proteins GRA6 and GRA7 of N. hughesi and Neospora caninum. This information can be used to differentiate the two species and contribute to further understanding of the prevalence and biology of N. hughesi. The newly defined proteins of N. hughesi are referred to as NhGRA6 and NhGRA7 in keeping with the protocol for naming homologous proteins of the Apicomplexa. Genes of the two dense granule proteins of N. hughesi (isolate Nh-A1) and four different isolates of N. caninum were isolated via PCR and their DNA sequences were determined. Computer analysis indicated that the two gene sequences were identical among all four N. caninum isolates. However, the gene for NhGRA6 was found to be 96 nucleotides longer at the 3' end than that of NcGRA6, resulting in a protein product that is 32 amino acids larger than NcGRA6. Two tandem repeat sequences were identified at the 3' end of the NhGRA6 gene. These repeat sequences contributed to the lengthening of the carboxy terminus of NhGRA6 in comparison with that of NcGRA6. The larger size of NhGRA6 was further confirmed by Western blot analysis in which NcGRA6 monospecific antibodies recognised a protein of approximately 42 kDa in N. hughesi whole **tachyzoite** preparation but a protein of 37 kDa in N. caninum whole **tachyzoite** preparation. Analysis of GRA7 gene sequences indicated a 6 and 14.8% difference at nucleotide and amino acid sequence level, respectively, between NcGRA7 and NhGRA7. Despite the same number of residues in the deduced amino acid sequences of all the GRA7 proteins, Western blot analysis indicated a difference in the migration pattern of NhGRA7 in comparison with NcGRA7. Results of our study indicate that diagnostic tests based on differences in dense granule sequences and antigenicity may have potential to differentiate between N. hughesi and N. caninum. Such diagnostic tests would be valuable tools to aid in our understanding of the epidemiology of these parasites. Additionally, dense granule proteins are **immunogenic** and they may have potential as use in recombinant vaccines against neosporosis.

L47 ANSWER 3 OF 8 MEDLINE DUPLICATE 2
2001354018 Document Number: 21127318. PubMed ID: 11223200.
Immunoconversion against **Sarcocystis neurona** in normal and dexamethasone-treated horses challenged with S. neurona sporocysts.
Cutler T J; MacKay R J; Ginn P E; Gillis K; Tanhauser S M; LeRay E V; Dame J B; Greiner E C. (Department of Pathobiology, PO Box 100880, College of Veterinary Medicine, University of Florida, Gainesville 32610, USA.)
VETERINARY PARASITOLOGY, (2001 Feb 26) 95 (2-4) 197-210. Journal code: XBU; 7602745. ISSN: 0304-4017. Pub. country: Netherlands. Language: English.

AB **Equine protozoal myeloencephalitis** is a common neurologic disease of horses in the Americas usually caused by **Sarcocystis neurona**. To date, the disease has not been induced in horses using characterized sporocysts from Didelphis

virginiana, the definitive host. S. neurona sporocysts from 15 naturally infected opossums were fed to horses seronegative for antibodies against S. neurona. Eight horses were given 5x10(5) sporocysts daily for 7 days. Horses were examined for abnormal clinical signs, and blood and cerebrospinal fluid were harvested at intervals for 90 days after the first day of challenge and analyzed both qualitatively (western blot) and quantitatively (anti-17kDa) for anti-S. neurona IgG. Four of the challenged horses were given dexamethasone (0.1mg/kg orally once daily) for the duration of the experiment. All challenged horses immunoconverted against S. neurona in blood within 32 days of challenge and in CSF within 61 days. There was a trend (P = 0.057) for horses given dexamethasone to immunoconvert earlier than horses that were not immunosuppressed. Anti-17kDa was detected in the CSF of all challenged horses by day 61. This response was statistically greater at day 32 in horses given dexamethasone. Control horses remained seronegative throughout the period in which all challenged horses converted. One control horse immunoconverted in blood at day 75 and in CSF at day 89. Signs of neurologic disease were mild to equivocal in challenged horses. Horses given dexamethasone had more severe signs of limb weakness than did horses
not given dexamethasone; however, we could not determine whether these signs were due to spinal cord disease or to effects of systemic illness. At necropsy, mild-moderate multifocal gliosis and neurophagia were found histologically in the spinal cords of 7/8 challenged horses. No organisms were seen either in routinely processed sections or by immunohistochemistry. Although neurologic disease comparable to naturally occurring **equine protozoal myeloencephalitis** (**EPM**) was not produced, we had clear evidence of an **immune response** to challenge both systemically and in the CNS. Broad immunosuppression with dexamethasone did not increase the severity of histologic changes in the CNS of challenged horses. Future work must focus on defining the factors that govern progression of inapparent S. neurona infection to **EPM**.

L47 ANSWER 4 OF 8 MEDLINE
2001354017 Document Number: 21127317. PubMed ID: 11223199.
Interpretation
of the detection of **Sarcocystis neurona** antibodies in the serum of young horses. Cook A G; Buechner-Maxwell V; Morrow J K; Ward D L; Parker N A; Dascanio J J; Ley W B; Cooper W. (Department of Large Animal Clinical Sciences, Virginia-Maryland College of Veterinary Medicine, Duck Pond Drive Phase II, Blacksburg, VA 24061, USA.. ancook2@vt.edu) . VETERINARY PARASITOLOGY, (2001 Feb 26) 95 (2-4) 187-95. Journal code: XBU; 7602745. ISSN: 0304-4017. Pub. country: Netherlands. Language: English.
AB Horses that are exposed to **Sarcocystis neurona**, a causative agent of **equine protozoal myeloencephalitis**, produce antibodies that are detectable in serum by western blot (WB). A positive test is indicative of exposure to the organism. Positive tests in young horses can be complicated by the presence of maternal antibodies. Passive transfer of maternal antibodies to S. neurona from seropositive mares to their foals was evaluated. Foals were sampled at birth (presuckle), at 24h of age (postsuckle), and at monthly intervals. All foals sampled before suckling were seronegative. Thirty-three foals from 33 seropositive mares became seropositive with

colostrum ingestion at 24h of age, confirming that passive transfer of S. neurona maternal antibodies occurs. Thirty-one of the 33 foals became seronegative by 9 months of age, with a mean seronegative conversion time of 4.2 months. These results indicate that evaluation of exposure to S. neurona by WB analysis of serum may be misleading in young horses.

L47 ANSWER 5 OF 8 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
2001075066 EMBASE A review of **Sarcocystis neurona** and **equine protozoal myeloencephalitis** (**EPM**). Dubey J.P.; Lindsay D.S.; Saville W.J.A.; Reed S.M.; Granstrom D.E.; Speer C.A.. J.P. Dubey, United States Dept. of Agriculture, Animal and Natural Resources Inst., Beltsville Agricultural Res. Center, Beltsville, MD 20705-2350, United States. jdubey@anri.barc.usda.gov. Veterinary Parasitology 95/2-4 (89-131) 26 Feb 2001.
Refs: 148.
ISSN: 0304-4017. CODEN: VPARDI.
Publisher Ident.: S 0304-4017(00)00384-8. Pub. Country: Netherlands.
Language: English. Summary Language: English.
AB **Equine protozoal myeloencephalitis** (**EPM**) is a serious neurological disease of horses in the Americas. The protozoan most commonly associated with **EPM** is **Sarcocystis neurona**. The complete life cycle of S. neurona is unknown, including its natural intermediate host that harbors its sarcocyst. Opossums (Didelphis virginiana, Didelphis albiventris) are its definitive hosts. Horses are considered its aberrant hosts because only schizonts and **merozoites** (no sarcocysts) are found in horses. **EPM**-like disease occurs in a variety of mammals including cats, mink, raccoons, skunks, Pacific harbor seals, ponies, and Southern sea otters. Cats can act as an experimental intermediate host harboring the sarcocyst stage after ingesting sporocysts. This paper reviews information on the history, structure, life cycle, biology, pathogenesis, induction of disease in animals, clinical signs, diagnosis, pathology, epidemiology, and treatment of **EPM** caused by S. neurona.

L47 ANSWER 6 OF 8 MEDLINE
2001156688 Document Number: 21088454. PubMed ID: 11219340. **Equine protozoal myeloencephalitis**. MacKay R J; Granstrom D E; Saville W J; Reed S M. (Department of Large Animal Clinical Sciences, College of Veterinary Medicine, University of Florida, Gainesville, Florida, USA.) VETERINARY CLINICS OF NORTH AMERICA. EQUINE PRACTICE, (2000 Dec) 16 (3) 405-25. Ref: 96. Journal code: CEP; 8511904. ISSN: 0749-0739. Pub. country: United States. Language: English.
AB Recent advances in the understanding of the parasite life cycle, epidemiology, clinical signs, diagnosis, treatment, and prevention of **EPM** are reviewed. The NAHMS Equine '98 study and a controlled retrospective study from The Ohio State University College of Veterinary Medicine identified a number of risk factors associated with development of the disease. The national annual incidence of **EPM** was 1% or less depending on the primary use of the animals. Increased disease risk was associated with age (1-5 and > 13 years of age), season (lowest in winter months and increasing with ambient temperature), previous stressful events, the presence of opossums, the use of nonsurface water drinking

systems, and failure to restrict wildlife access to feed. Horses that received treatment were 10 times more likely to improve, and those that improved were 50 times more likely to survive. A number of recent studies confirmed that horses can be experimentally infected with S. neurona; however, large numbers of sporocysts are apparently necessary to achieve infection, and clinical signs and abnormal CNS histology are only seen inconsistently. Results suggest that CNS infection and positive CSF immunoblot findings may be transient phenomena among naturally infected horses. Although immunosuppression may be involved in the development of **EPM**, some element of the **immune response** seems to be necessary for the development of clinical signs. Use of the standard immunoblot test for the detection of anti-S. neurona antibodies in CSF continues to provide the most useful adjunct to a detailed neurologic examination for the diagnosis of **EPM**. Test sensitivity and specificity were 89% in 295 horses euthanatized because of neurologic disease, of which 123 were confirmed cases of **EPM**. The PPV was 85%, and the NVP was 92%. A number of promising new **EPM** treatments are under investigation. In addition to standard SDZ/PYR therapy, toltrazuril, ponazuril, diclazuril, and NTZ have shown promise as possible alternatives.

L47 ANSWER 7 OF 8 WPIDS COPYRIGHT 2001 DERWENT INFORMATION LTD
AN 1999-571872 [48] WPIDS
AB WO 9947927 A UPAB: 19991122
NOVELTY - Biologically pure culture of equine Neospora, is new.
DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:
(a) detecting antibodies (Ab) specifically reactive with equine Neospora antigens (Ag) by forming an Ab-Ag complex;
(b) detecting Neospora by forming a complex with an antibody (Ab1) specifically reactive with Neospora antigen;
(c) detecting Neospora-specific nucleic acid (I) by hybridization with a specific oligonucleotide probe; and
(d) pharmaceutical composition containing equine Neospora **immunogen** and a carrier.
ACTIVITY - Antiprotozoal.
MECHANISM OF ACTION - Induction of a specific **immune response**.
USE - **Immunogens** (optionally expressed from gene therapy vectors) from equine Neospora are used in vaccines for treatment or prevention of Neospora infection in horses and other animals. Neospora is a causative agent of **equine protozoal myeloencephalitis** (**EPM**). Detection of Neospora-specific antigens, antibodies or nucleic acid (by usual immunoassay or hybridization tests) is used to diagnose infection. Antibodies (Ab) specific for equine Neospora are used for diagnosis; to select candidate **immunogens** for vaccine development; to isolate proteins; to screen DNA libraries and as therapeutic/prophylactic agents.
ADVANTAGE - Reagents specific for equine Neospora allow differentiation between **equine protozoal myeloencephalitis** caused by Neospora and **Sarcocystis neurona**. These pathogens require different treatments and treatment of Neospora is only effective if applied before the parasite has

formed cysts. The vaccines also prevent shedding of oocysts by animals known to be infected.
Dwg.0/2

L47 ANSWER 8 OF 8 MEDLINE DUPLICATE 3
1998234002 Document Number: 98234002. PubMed ID: 9573058. Evidence that surface proteins Sn14 and Sn16 of **Sarcocystis neurona merozoites** are involved in infection and **immunity**. Liang F T; Granstrom D E; Zhao X M; Timoney J F. (Gluck Equine Research Center, Department of Veterinary Science, University of Kentucky, Lexington 40546-0099, USA.) INFECTION AND IMMUNITY, (1998 May) 66 (5) 1834-8. Journal code: GO7; 0246127. ISSN: 0019-9567. Pub. country: United States. Language: English.

AB **Sarcocystis neurona** is the etiologic agent of **equine protozoal myeloencephalitis** (**EPM**). Based on an analysis of 25,000 equine serum and cerebrospinal fluid (CSF) samples, including samples from horses with neurologic signs typical of **EPM** or with histologically or parasitologically confirmed **EPM**, four major immunoblot band patterns have been identified. Twenty-three serum and CSF samples representing each of the four immunoblot patterns were selected from 220 samples from horses with neurologic signs resembling **EPM** and examined for inhibitory effects on the infectivity of S. neurona by an in vitro neutralization assay. A high correlation between immunoblot band pattern and neutralizing activity was detected. Two proteins, Sn14 and Sn16 (14 and 16 kDa, respectively), appeared to be important for in vitro infection. A combination of the results of surface protein labeling, immunoprecipitation, Western blotting, and trypsin digestion suggests that these molecules are surface proteins and may be useful components of a vaccine against S. neurona infection. Although S. neurona is an obligate intracellular parasite, it is potentially a target for specific antibodies which may lyse **merozoites** via complement or inhibit their attachment and penetration to host cells.

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whalen, j?/au,in or whalen j?/au,in
'IN' IS NOT A VALID FIELD CODE
L48           2 FILE MEDLINE
L49           0 FILE CAPLUS
L50           2 FILE BIOSIS
'IN' IS NOT A VALID FIELD CODE
L51           0 FILE EMBASE
L52           0 FILE WPIDS
L53           0 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L54           4 BIGBIE, R?/AU,IN OR BIGBIE R?/AU,IN


'IN' IS NOT A VALID FIELD CODE
'AUIN' IS NOT A VALID FIELD CODE
L55         584 FILE MEDLINE
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L57          746 FILE BIOSIS
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'AUIN' IS NOT A VALID FIELD CODE
L58          509 FILE EMBASE
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L59           62 FILE WPIDS
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L60            6 FILE JICST-EPLUS

TOTAL FOR ALL FILES
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L63          272 FILE CAPLUS
L64          260 FILE BIOSIS
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L65          166 FILE EMBASE
L66           31 FILE WPIDS
L67            0 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L68          948 WHALEN, J?/AU,IN OR WHALEN J?/AU,IN

=> s l54 and l61 and l68
L69            0 FILE MEDLINE
L70            0 FILE CAPLUS
L71            0 FILE BIOSIS
L72            0 FILE EMBASE
L73            0 FILE WPIDS
L74            0 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L75            0 L54 AND L61 AND L68

=> s (l54 or l61 or l68) and (l10 or l17 or l24)
L76            0 FILE MEDLINE
L77            0 FILE CAPLUS
L78            0 FILE BIOSIS
L79            0 FILE EMBASE
L80            0 FILE WPIDS
L81            0 FILE JICST-EPLUS

TOTAL FOR ALL FILES
L82            0 (L54 OR L61 OR L68) AND (L10 OR L17 OR L24)

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COST IN U.S. DOLLARS                          SINCE FILE      TOTAL
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